

Attorney Docket No. 3817.14-1
Customer No. 23308

Amendments to the Claims:

This listing of claims will replace all prior versions, and listing, of claims in the application:

Listing of Claims:

1. (Currently Amended): A protein binding assay for measuring inositol 1,4,5-triphosphate (IP₃) in a sample employing as reagents a conjugate of IP₃, a detectable fluorescent label joined through - R - Z -, wherein R is a bond or linker at the 2-hydroxyl position of said IP₃, R is of not more than 16 atoms in the chain and Z is selected from the group consisting of oxy, amino, thio, succinimidyl, amino, ureido, ester, phospho, thiophospho, and oxalo, and as a binding protein a 226 – 578 amino acid extracellular portion of mouse inositol 1,4,5-triphosphate receptor (IP₃R) having at least about 200 times the affinity for IP₃ than the intact IP₃R, wherein said conjugate and IP₃ in the sample compete for binding to said binding protein and the amount of bound or unbound conjugate will be related to the number of binding proteins bound by IP₃ in said sample, said method comprising:

combining in an assay medium said sample, said conjugate and said binding protein and incubating said mixture for sufficient time for complex formation of IP₃ and said conjugate with said binding protein; and

detecting the bound or unbound label as a measure of the IP₃ present in the sample.
2. (Original): A protein binding assay according to Claim 1, wherein said assay is in a homogeneous format.

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3. (Original): A protein binding assay according to Claim 1, wherein said sample is a cellular lysate, and wherein said cellular lysate has been treated to block kinases and phosphatases and prepare said sample for said assay.
4. (Previously Presented): A protein binding assay according to Claim 1, wherein said binding protein is of not more than about 600 amino acids and comprises at least amino acids 226 – 578 of the mouse IP₃R Type 1 fused to glutathione-S-transferase.
5. (Canceled).
6. (Original): A protein binding assay according to Claim 1, wherein said binding protein is a fusion protein of up to about 1.5kD amino acids.
7. (Currently Amended): A protein binding assay according to Claim 1, wherein said label is ~~a fluorescer~~ 2-O- (2-aminoethyl- (6-carboxamidofluoresceinyl)).
8. (Original): A method according to Claim 1, wherein the order of addition of reagents is: (a) combining said sample with said binding protein; and (b) adding said conjugate, with incubating after (a) and (b).
- 9 -21. (Canceled).